Remarks

Claims 11-14, 16-18, and 20-23 are pending in the current application. Applicant has amended Claim 11 to incorporate the subject matter of Claim 19 and 27 therein. Accordingly, Applicants have canceled Claims 19 and 27 without disclaiming the subject matter contained therein. Further, the Applicant has amended Claims 20 and 21 to provide the term "concentration" with a proper antecedent basis. Support for this amendment can be found in paragraphs 35-42. Further, the Applicant has added new Claim 28. Support for Claim 28 can be found in paragraphs 24-28.

Claim Rejections – 35 U.S.C. § 112

Claim 19 has been rejected for lack of antecedent basis for the term "concentration (C)." The Applicant has canceled Claim 19 and as a result the rejection is now moot. The Applicants note that Claims 20 and 21 have been amended to provide that the DNA compaction agent "has a concentration (C) that is defined by the following equation." Thus, the Applicants have provided the term concentration in both Claims 20 and 21 with a proper antecedent basis. In light of the phone conversation between the Examiner and the Applicant's representative on September 28, 2004 the erroneous rejection of the claims based on improper numbering is now moot. Accordingly, the Applicant requests withdrawal of this rejection and submits that the claims are numbered in accordance with 37 CFR 1.126.

Claims Rejections – 35 U.S.C. § 102

Claim 11-14, 16-18, 22, 23 and 27 have been rejected under 35 U.S.C. § 102(b) as anticipated by Hodgson (U.S. Patent No. 6,410,220). The Applicants submit that as a result of the amendment to Claim 11 to incorporate the subject matter of Claims 19 and 27, the rejection is now obviated. In particular, nothing in Hodgson discloses a DNA compaction agent that is present during

a ligation reaction and at a concentration sufficient to make a DNA insert flexible so that the DNA insert can ligate with a DNA vector to make a circularized nucleic acid molecule.

Hodgson generally describes a method of creating recombinant molecules through self-ligating and self-assembly of DNA fragments having cohesive ends. In particular, Hodgson discloses the use of a DNA condensing agent to protect DNA from shearing. The DNA condensing agent is applied **after** two or more fragments of DNA have been ligated together and are applied directly to the ligation medium. The Examiner's attention is invited to column 23, lines 41-49 wherein Hodgson states that:

> **It is also desirable to avoid shearing the DNA once large segments have been joined by ligation.** One method of avoiding shear is to add the transfection agent, such as Superfect™ reagent (dendrimers, Qiagen) or Lipofectamine™ (Liposomes, Life Technologies, Gaithersburg, Md.) diredtly to the ligation reaction, and then add the cells to be transfected to the mixture. This, or a similar method avoids the need to physically move the ligated DNA, and thus prevents shearing. Another method is to add a DNA condensing reagent (dendrimers, polycations [such as polyethyleneamine] histones or liposomes) directly to the DNA ligation reaction, and then move the DNA by pipette after it has condensed (thus reducing shearing of the DNA). Once inside the cell, viral DNA can replicate (as in the examples of partially replication-competent adenovirus and herpes simplex virus vectors). [Emphasis added].

Consequently, Hodgson discloses adding a DNA condensing agent after ligation in order to reduce DNA shearing. In contrast, the Applicant claims "producing circularized recombinant nucleic acid by ligating a DNA insert and DNA vector in the presence of a DNA compaction agent." Accordingly, the Applicants respectfully request withdrawal of the rejection of Claims 11-14, 16-18, 22, and 23 as anticipated by Hodgson.

Claims Rejections Under 35 U.S.C. § 103

Claims 19-21 have been rejected under 35 U.S.C. § 103(a) as being unpatentable over

Hodgson in view of Nagaki (BBRC 246:137-141, 1998). Claim 19 has been canceled and as a result a rejection of this claim is now moot. Nevertheless, to the extent that the subject matter of Claim 19 has been incorporated into Claim 11, the Applicant responds accordingly. It must be kept in mind under the appropriate test of obviousness under 35 U.S.C. §103 that:

> Where claimed subject matter has been rejected as obvious in view of a combination of prior art references, a proper analysis under §103 requires, inter alia, consideration of two factors: (1) whether the prior art would have suggested to those of ordinary skill in the art that they should make the claimed composition or device, or carry out the claimed process; and (2) whether the prior art would also have revealed that in so making or carrying out, those of ordinary skill would have a reasonable expectation of success. See In re Dow Chemical Co., 837 F.2d 469, 473, 5 U.S.P.Q. 2d 1529, 1531 (Fed.Cir. 1988). Both the suggestion and the reasonable expectation of success must be founded in the prior art, not in the applicant's disclosure. In re Vaeck, 20 U.S.P.Q. 2d 1438, 1442 (Fed. Cir. 1991).

Hodgson teaches adding a DNA condensing agent **after** ligation in order to reduce DNA shearing. In contrast, the Applicants ligate a DNA insert to a DNA vector in the presence of a compaction agent. The use of the compaction agent during the ligation process provides the Applicant's DNA insert with enough flexibility to facilitate the construction of large circularized recombinant nucleic acid molecules, for example, that are greater than 5kb. Nothing in Hodgson discloses or suggests the use of a DNA compacting agent to provide flexibility to a DNA insert in order to facilitate the ligation of a large DNA insert into a DNA vector. Rather, Hodgson discloses a method to reduce DNA shearing by applying a compaction agent after ligation has occurred. Thus, nothing in Hodgson suggests modifying the technique described therein, because the sole purpose of the DNA compaction agent in Hodgson is to prevent DNA shearing after ligation had already occurred. Thus, there is no suggestion or teaching in Hodgson to add a DNA compaction agent as part of the ligation process.

Nagaki teaches the use of HMG1 and 2 to help facilitate double stranded break (DSB) repair of a short, (3,162 bp) DNA molecule (pUC119 Vector), which is linearized prior to the application

of HMG. Importantly, Nagaki only shows the production of linear and circular monomers of pUC119, and linear dimers which are not circularized. Therefore, the Nagaki method only results in the formation of a **circular monomer** (i.e. a vector without an insert). Nagaki never achieved the formation of circularized dimers (i.e. a vector with an insert). Specifically, the Nagaki method teaches the use of HMG1 and 2 **for intra-molecular** ligation to create short sized circular vector monomers, which do not contain a DNA insert. The Applicant invites the Examiner to page 140, right column of Nagaki, which states that "cohesive-ended pUC119 DNA was ligated by DNA ligase IV, resulting in circular DNA by –intra-molecular ligation and linear products by inter-molecular ligation." The Examiner's attention is invited to Fig. 2-5, which demonstrate that HMG 1 and 2 only resulted in circular monomers, and linear monomers and dimers.

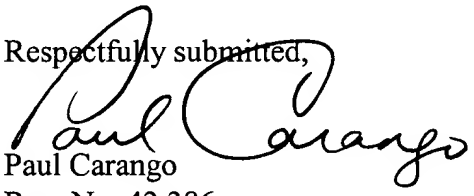
Nagaki does not teach or suggest that a DNA insert can be ligated into a DNA vector to form a recombinant nucleic acid molecule that is 5kb or greater. In fact, Nagaki shows that the use of HMG 1 and 2 does not result in the claimed circularized recombinant DNA with a DNA insert. As is well known in the art, an intra-molecular ligation to form a circular monomer is different from an inter-molecular ligation of a DNA insert and a DNA vector which forms a recombinant circularized nucleic acid. In fact, one skilled in the art would understand that intra-molecular ligation is energetically favored over inter-molecular ligation. One skilled in the art would also understand that DNA which is highly concentrated favors inter-molecular ligation, whereas DNA that is diluted in solution favors intra-molecular ligation. One skilled in the art would therefore have no reasonable expectation of success from the teaching of Nagaki that HMG could function to ligate DNA inserts into a vector. In fact, Nagaki teaches away from using HMG to ligate a DNA insert into a DNA vector, since Nagaki failed to form circularized dimers.

Nothing in Nagaki suggests to one skilled in the art that Hodgson should be modified to

include HMG 1 and 2 in the ligation reaction as ligation proceeds, or that HMG 1 and 2 could be used to facilitate the integration of a DNA insert into a DNA vector. One skilled in the art would have no reasonable expectation of success in achieving the Applicant's claimed invention from the combination of Nagaki and Hodgson, because Hodgson shows that adding compaction agent after ligation reduces shearing, while Nagaki's results demonstrate that compaction agent did not result in the claimed vectors with inserts. In light of the shortcomings in both Nagaki and Hodgson, one skilled in the art would not have any reasonable expectation that HMG would facilitate a DNA ligation reaction which ligates a DNA insert into a DNA vector. Accordingly, the Applicant respectfully requests withdrawal of the rejection of Claims 19-21 as unpatentable over Hodgson in view of Nagaki.

The Applicants submit that the Application is now in condition of allowance, which is respectfully requested.

Respectfully submitted,



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